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CLONING OF *XRCC2* AND *XRCC3*, HUMAN HOMOLOGS OF THE *RAD51* DNA REPAIR GENE. L.H. Thompson, N. Liu*, R. S. Tebbs*, J. E. Lamerdin*, J.D. Tucker, and A. V. Carrano, Biology and Biotechnology Research Program, Lawrence Livermore National Laboratory, P.O. Box 808, Livermore, CA 94551.

To delineate DNA repair pathways that act on damage from ionizing radiation and DNA cross-linking agents, we have identified human genes that correct repair deficient rodent cell mutants. The mutants irs1 and irs1SF were isolated as x-ray-sensitive clones from V79 and CHO cell lines, respectively. Human genes that correct their extreme mitomycin C sensitivities (~60-fold) were identified and mapped by analyzing somatic cell hybrids. To clone these genes, the mutants were transfected with the pEBS7 cDNA expression libraries kindly provided by Randy Legerski. A functional *XRCC3* cDNA was obtained from a secondary transformant of irs1SF by screening a cosmid library with the hygromycin gene. The resulting cDNA only partially corrected the mutagen sensitivities of irs1SF but efficiently corrected its high chromosomal instability. One pEBS7 library transformant of irs1 was very unstable, suggesting correction by an episomally replicating plasmid. A functional cDNA of *XRCC2* was rescued from a Hirt extract of this primary transformant. The *XRCC2* cDNA efficiently corrected the MMC, cisplatin, and EMS sensitivities of irs1, and stable transformants were obtained by transfecting the cDNA carried in the pcDNA3 expression vector. Sequence analysis revealed that both *XRCC2* and *XRCC3* are distant homologs of *RAD51*, a gene required for meiotic recombination and double-strand break repair in the yeast *S. cerevisiae*. Highly conserved mammalian homologs of RAD51 have already been reported, with the human homolog (HHR51) showing 57% identify with RAD51. HHR51 is an analog of the bacterial RecA protein, which performs homologous pairing and strand exchange during recombination. The open reading frames of *XRCC2* and *XRCC3* encode proteins of 280 a.a. and 346 a.a., respectively. Alignment of the 240 a.a. C-terminal region of XRCC2 shows 19% identity with RAD51, and the 246 a.a. C-terminal region of XRCC3 shows 27% identity with RAD51. These similarities suggest that the XRCC2 and XRCC3 proteins may participate in a recombinational repair pathway that efficiently removes DNA cross-links. (Work done under the auspices of the U.S. DOE by LLNL under contract No. W-7405-ENG-48.)